CTACTGAGGACCACAAAATGGTAGAAGCCTTACGTGTTTGGGCATCTACTCATATGTCACCGTCTTGGACATTACTAAAA TTGTGTGATGTTCAGCCAATGCAGTATTTTGACCTGACTTGTCAGCTCTTGGGCAAAGCAGAAGTGGACGGAGCATCATT TCTTCTAAAGGTATGGGATGGCACCAGGACACCATTTCCATCTTGGAGAGTCTTAATACAAGACCTTGTTCTTGAAGGTG ATTTAAGTCACATCCATCGGCTACAAAATCTGACAATAGACATTTTAGTCTACGATAACCATGTTCATGTGGCAAGATCT AAGTTTAGAGTTTCATCTTCATGGAGGTACCAGTTACGGTCGGGGAATCAGGGTCTTACCAGAAAGTAACTCTGATGTGG **ATCAACTGAAAAAGGATTTAGAATCTGCAAATTTGACAGCCAATCAGCATTCAGATGTTATCTGTCAATCAGAACCTGAC** GACAGCTTTCCAAGCTCTGGATCAGTATCATTATACGAGGTAGAAAGATGTCAACAGCTATCTGCTACAATACTTACAGA **TCATCAGTATTTGGAGAGGACACCACTATGTGCCATTTTGAAACAAAAGCTCCTCAACAATACCGCATCCGAGCAAAAT** TGAGGTCATATAAGCCCAGAAGACTATTTCAGTCTGTTAAACTTCATTGCCCTAAATGTCATTTGCTGCAAGAAGTTCCA CATGAGGGCGATTTGGATATAATTTTTCAGGATGGTGCAACTAAAACCCCAGATGTCAAGCTACAAAATACATCATTATA TGATTCAAAAATCTGGACCACTAAAAATCAAAAAGGACGAAAAGTAGCAGTTCATTTTGTGAAAAATAATGGTATTCTCC CGCTTTCAAATGAATGTCTACTTTTGATAGAAGGAGGTACACTCAGTGAAATTTGCAAACTCTCGAACAAGTTTAATAGT GTAATTCCTGTGAGATCTGGCCACGAAGACCTGGAACTTTTGGACCTTTCAGCACCATTTCTTATACAAGGAACAATACA TCACTATGGATGTAAACAGTGTTCTAGTTTGAGATCCATACAAAATCTAAATTCCCTGGTTGATAAAACATCGTGGATTC TCAGAAAAGTGTGGATATGATCATGGATATGTTTTGTCCTCCAGGAATAAAAATTGATGCATATCCGTGGTTGGAATGCT TCATCAAGTCATACAATGTCACAAATGGAACAGATAATCAAATTTGCTATCAGATTTTTGACACCACAGTTGCAGAAGAT GTAATCGTACGTAGGAATTCTAGATCTatgGACTACAAAGACGATGACGACAAGatgCCGCGCGCCCCCCCGCTGCCGAGC CGTGCGCTCCTGCTGCGCAGCCACTACCGCGAGGTGCTGCCGCTGGCCACGTTCGTGCGGCGCCTGGGGCCCCAGGGCT GGCGCTGGTGCAGCGGGGACCCGGCGGCTTTCCGCGGCGCTGGTGCCCAGTGCCTGGTGCCCTGGGACGCA CGAGCGCGCGCGAAGAACGTGCTGGCCTTCGGCCTTCGCGCTGCTGGACGGGGCCCGGGGGGCCCCCGAGGCCTTCA CGCGTGGGCGACGTGCTGGTTCACCTGCTGGCACGCTGCGCGCTCTTTGTGCTGGTGGCTCCCAGCTGCGCCTACCA GTCTGGGATGCGAACGGCCTGGAACCATAGCGTCAGGGAGGCCGGGGTCCCCCTGGGCCTGCCAGCCCCGGGTGCGAGG AGGCGCGGGGCAGTGCCAcCGAAGTCTGCCGTTGCCCAAGAGGCCCAGGCGTGGCGCTGCCCCTGAGCCGGAGCGGAC GCCCGTTGGGCAGGGGTCCTGGGCCCACCCGGGCAGGACGCGTGGACCGAGTGACCGTGGTTTCTGTGTGTCACCTG CACCACGCGGGCCCCCCATCCACATCGCGGCCACCACGTCCCTGGGACACGCCTTGTCCCCCGGTGTACGCCGAGACCAA GCGCTCGGAGGCTCGTGGAGACCATCTTTCTGGGTTCCAGGCCCTGGATGCCAGGGACTCCCCGCAGGTTGCCCCGCCTG $\tt CCCCAGCGCTACTGGCAAATGCGGCCCCTGTTTCTGGAGCTGCTTGGGAACCACGCGCAGTGCCCCTACGGGGTGCTCCT$ CAAGACGCACTGCCCGCTGCGAGCTGCGGTCACCCCAGCAGCCGGTGTCTGTGCCCGGGAGAAGCCCCAGGGCTCTGTGG CGGCCCCGAGGAGGAGGACACAGACCCCCGTCGCCTGGTGCAGCTGCTCCGCCAGCACAGCAGCCCCCTGGCAGGTGTAC GAACACCAAGAAGTTCATCTCCCTGGGGAAGCATGCCAAGCTCTCGCTGCAGGAGCTGACGTGGAAGATGAGCGTGCGGG GCTGCGCTTGGCTGCGCAGGAGCCCAGGGGTTGGCTGTGTTCCGGCCGCAGAGCACCGTCTGCGTGAGGAGATCCTGGCC **AAAGAACAGGCTCTTTTTCTACCGGAAGAGTGTCTGGAGCAAGTTGCAAAGCATTGGAATCAGACAGCACTTGAAGAGGG** TTCATCCCCAAGCCTGACGGGCTGCGGCCGATTGTGAACATGGACTACGTCGTGGGAGCCAGAACGTTCCGCAGAGAAAA GAGGGCCGAGCGTCTCACCTCGAGGGTGAAGGCACTGTTCAGCGTGCTCAACTACGAGCGGGCGCGGCGCCCCCGGCCTCC TGGGCGCCTCTGTGCTGGGCCTGGACGATATCCACAGGGCCTGGCGCACCTTCGTGCTGCTGCGGGCCCAGGACCCG $CCGCCTGAGCTGTACTTTGTCAAGGTGGATGTGACGGGCGCGTACGACACCATCCCCCA<math>\sigma$ GACAGGCTCACGGAGGTCAT GCAAGGCCTTCAAGAGCCACGTCTCTACCTTGACAGACCTCCAGCCGTACATGCGACAGTTCGTGGCTCACCTGCAGGAG ACCAGCCGCTGAGGGATGCCGTCGTCATCGAGCAGAGCTCCTCCCTGAATGAGGCCAGCAGTGGCCTCTTCGACGTCTT CCTACGCTTCATGTGCCACCACGCCGTGCGCATCAGGGGGCAAGTCCTACGTCCAGTGCCAGGGGATCCCGCAGGGCTCCA TCCTCTCCACGCTGCTCTGCAGCCTGTGCTACGGCGACATGGAGAACAAGCTGTTTGCGGGGGATTCGGCGGGACGGGCTG CTCCTGCGTTTGGTGGATGATTTCTTGTTGGTGACACCTCACCTCACCCCACGCGAAAACCTTCCTCAGGACCCTGGTCCG AGGTGTCCCTGAGTATGGCTGCGTGAACTTGCGGAAGACAGTGGTGAACTTCCCTGTAGAAGACGAGGCCCTGGGTG GCACGGCTTTTGTTCAGATGCCGGCCCACGGCCTATTCCCCTGGTGCGGCCTGCTGGATACCCGGACCCTGGAGGTG CAGAGCGACTACTCCAGCTATGCCCGGACCTCCATCAGAGCCAGTCTCACCTTCAACCGCGGCTTCAAGGCTGGGAGGAA CATGCGTCGCAAACTCTTTGGGGTCTTGCGGCTGAAGTGTCACAGCCTGTTTCTGGATTTGCAGGTGAACAGCCTCCAGA CGGTGTGCACCAACATCTACAAGATCCTCCTGCTGCAGGCGTACAGGTTTCACGCATGTGTGCTGCAGCTCCCATTTCAT CAGCAAGTTTGGAAGAACCCCACATTTTTCCTGCGCGTCATCTCTGACACGGCCTCCCTGCTACTCCATCCTGAAAGC CAAGAACGCAGGGATGTCGCTGGGGGCCAAGGGCGCCCCCCGGCCCTCTGCCCTGCGAGGCCGTGCAGTGCCTGTGCCACC **AAGCATTCCTGCTCAAGCTGACTCGACACCGTGTCACCTACGTGCCACTCCTGGGGTCACTCAGGACAGCCCAGACGCCA** CTGAGTCGGAAGCTCCCGGGGACGACGCTGACTGCCCTGGAGGCCGCAGCCAACCCGGCACTGCCCTCAGACTTCAAGAC CATCCTGGACTGAGTCGAC

Figure 1. Nucleotide sequence of encoded hPot1-hTERT fusion protein.

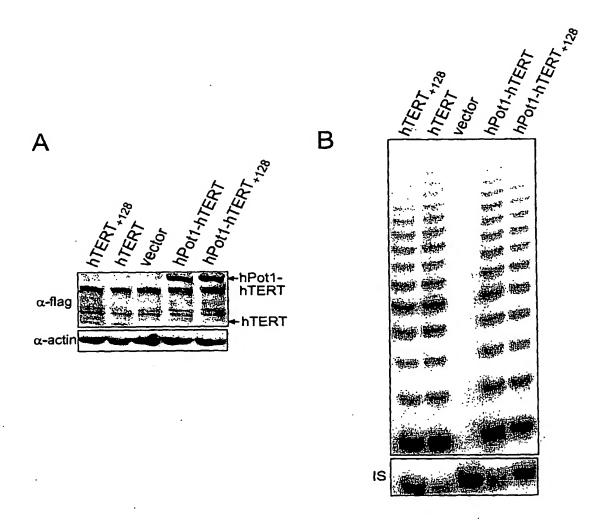


Figure 2. hTERT proteins retain telomerase activity when fused to hPot1. (A) Lysates from HA5 cell expressing the described constructs were immunoblotted with anti-flag antibodies to detect ectopic hTERT containing proteins. Actin serves as a loading control. (B) Lysates were assayed for in vitro telomerase activity. The internal standard (IS) served as a positive control for PCR amplification.

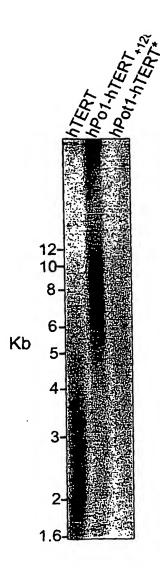


Figure 3. The hPot1-hTERT chimeric proteins elongate telomeres. Restriction enzyme digested genomic DNA isolated from late passage HA5 cells expressing hTERT, hPot-hTERT or hPot1-hTERT+128 and from HA5 cells were hybridized with a telomeric probe to visualize telomere containing fragments. Left, molecular weight markers in kilo bases (Kb). Sample for hPot1-hTERT was underloaded (*).

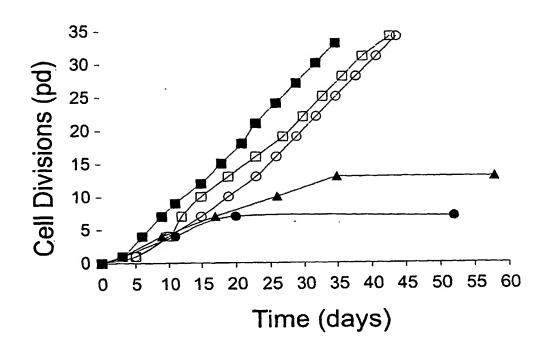


Figure 4. HA5 cells expressing hPot1-hTERT fusion proteins immortalize. The lifespan in population doublings (pd) of HA5 cell lines infected with vectors expressing hPot1-hTERT (□), hPot1-hTERT+128 (○) or controls expressing vector alone (▲), hTERT (■) or hTERT+128 (●) is plotted against time in days.